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Isohydric regulation of plasma potassium by bicarbonate in the rat

DONALD S. FRALEY and SHELDON ADLER

Department of Medicine, University of Pittsburgh School of Medicine and Montefiore Hospital, Pittsburgh, Pennsylvania

Isohydric regulation of plasma potassium by bicarbonate in the rat. pH and bicarbonate affect many metabolic reactions but each may change independently. To study bicarbonate's effect on plasma potassium, blood bicarbonate in normal, hypokalemic or hyperkalemic rats was either maintained constant, lowered by hydrochloric acid or raised by sodium bicarbonate administration. Blood pH was maintained constant by changing P_{CO_2}. In normokalemia lowering bicarbonate increased plasma potassium 2.0 mEq above values obtained in the other groups. To eliminate urinary potassium losses, experiments were also performed in rats with bilateral ureteral ligation. Again, plasma potassium concentration rose significantly more in the lowered bicarbonate group. Similarly, in hypokalemia, plasma potassium rose 1.2 and 0.4 mEq in the lowered and unchanged groups, but fell 0.2 mEq/liter in the elevated group. Differences could not be ascribed to renal potassium losses as potassium excretion was essentially zero in each group. In hyperkalemia, plasma potassium concentration remained elevated for 150 min in the lowered bicarbonate group but fell 1.3 and 2.0 mEq in the unchanged and elevated groups, respectively. Urinary potassium losses in the three groups were statistically identical. In all experiments blood pH was maintained unchanged during the experiment. The data show that bicarbonate, independent of blood pH, alters transcellular potassium distribution suggesting the usefulness of bicarbonate therapy in hyperkalemia even at a compensated blood pH.

Régulation isohydrique du potassium plasmatique par le bicarbonate chez le rat. Le pH et le bicarbonate peuvent affecter de nombreuses réactions métaboliques mais chacun de ces facteurs peut varier indépendamment de l'autre. Afin d'étudier l'effet du bicarbonate sur le potassium plasmatique, le bicarbonate plasmatique a été maintenu constant, ou abaissé par l'acide chlorhydrique ou augmenté par le bicarbonate du sodium chez des rats normaux, ou hypokaliémiques, ou hyperkaliémiques. La constance du pH du sang était assurée par des modifications adéquates de la P_{CO_2}. En normokaliémie, l'abaissement du bicarbonate augmente la kaliémie de 2,0 mEq/litre par rapport aux valeurs obtenues dans les autres groupes. Afin d'éliminer les pertes urinaires de potassium ces expériences ont été réalisées aussi chez des rats dont les deux uretères étaient liés. A nouveau, le potassium plasmatique a augmenté significativement plus dans le groupe dont le bicarbonate était abaissé. En hypokaliémie, de la même façon, le potassium plasmatique augmente de 1,2 et 0,5 mEq/litre dans les groupes dont le bicarbonate est, respectivement abaissé ou normal, mais il baisse de 0,2 mEq/litre dans le groupe à bicarbonate élevé. Ces différences ne peuvent pas être mises sur le compte de pertes rénales de potassium car celles-ci ont été pratiquement nulles dans les divers groupes. En hyperkaliémie le potassium plasmatique reste élevé pendant 150 minutes dans le groupe dont le bicarbonate est abaissé mais le potassium baisse de 1,3 et 2,0 mEq/litre dans les groupes dont le bicarbonate est, respectivement, normal et élevé. Les pertes urinaires de potassium dans les trois groupes sont semblables. Dans toutes les expériences le pH sanguin a été maintenu à une valeur constante. Les résultats montrent que le bicarbonate, indépendamment du pH du sang, modifie la distribution transcellulaire du potassium, ce qui suggère l' utilité du traitement de l'hyperkaliémie par le bicarbonate même quand le pH sanguin est compensé.

Extracellular potassium represents less than 2% of total body potassium [1]. Relatively small changes in cellular compartment potassium can result, therefore, in large changes in plasma potassium concentration. Accordingly, plasma potassium concentration may be reduced, normal or elevated despite normal body potassium stores. A major regulator of transcellular potassium distribution is the pH of the extracellular fluid as reflected by the blood pH. Much evidence has accumulated showing that lowering blood pH raises serum potassium concentration while the converse occurs when blood pH is elevated [2-5]. Recently, it has been found that extracellular bicarbonate concentration, separate from its effect on extracellular pH, affects a wide range of metabolic reactions [6-9]. There is conflicting evidence, however, as to whether alteration in the blood bicarbonate concentration, under isohydric conditions, alters the plasma potassium concentration [10-13] in normokalemia and there is no information on bicarbonate's role in hypokalemia or hyperkalemia.

To study this problem, the blood bicarbonate concentration was altered in normokalemic, hypokalemic and hyperkalemic rats *in vivo* while arterial blood pH was held constant. The results show that extracellular bicarbonate concentration regulates plasma potassium concentration in each of the potassium states. As this effect is independent of both

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blood pH and urinary potassium excretion, it appear that bicarbonate alters the transcelllular distribution of potassium at all levels of plasma potassium concentration.

Methods

Intact male Sprague-Dawley rats weighting 350 to 500 g were anesthetized with Inactin, 100 mg/kg i.p. in all experiments. A tracheostomy was performed and a jugular venous catheter (P50) was inserted and attached to a Harvard infusion syringe pump. A polyethylene (P50) catheter was inserted in the femoral artery and connected to a heparinized syringe for blood sampling. A polyethylene catheter (P90) was inserted in the bladder through a suprapubic incision and connected to a collection tube. Temperature was monitored by thermometer rectally. The animal was placed on a perforated plywood board positioned on a heating pad to maintain a constant body temperature throughout the experiments. Unless otherwise noted, animals were infused with 2.1 ml of saline per hour through the venous catheter. The tracheostomy tube was attached to a Harvard Apparatus Rodent Respirator and the animals were ventilated with a gas mixture containing 30% oxygen and 70% nitrogen unless otherwise noted. After placement on the respirator, animals were given succinyl choline to prevent attempts at respiratory compensation. Ventilatory rate was adjusted to 120/min and tidal volume to 2 ml unless otherwise stated. In all experiments arterial blood samples of 0.8 ml were drawn at 30 and 60 min and acid-base indices measured to determine the presence of a steady state. If there were significant differences between these indices in the two time periods, the experiment was discontinued. This 60-min control period was employed in all experiments. A 150-min experimental period was then begun. Another 0.8-ml blood sample was obtained after 120 min (E_1). At the conclusion of the 150-min experimental period, rats were killed by exsanguination from the abdominal aorta into a heparinized syringe (E_2). Gluteal muscle tissue for potassium determination was obtained as previously described [4]. Experiments performed using this basic model are described in the following.

Effect of F_iO_2 on plasma potassium concentration. Rats were ventilated with room air for the 60-min control period and then divided into three groups. One group was ventilated with a 100% oxygen gas mixture, a second was ventilated with a 30% oxygen-70% nitrogen gas mixture, while the third group was continued on room air.

Respiratory acidosis experiments. At the conclusion of the control period, respiratory acidosis was induced by ventilating the animals with a 10% carbon dioxide, 30% oxygen, 60% nitrogen gas mixture. The rate and volume of ventilation were unchanged.

Isohydric experiments in normokalemic rats with intact renal excretory capacity. At the end of the control period, rats were divided into three groups. The first, or normal bicarbonate group, was ventilated with the 30% oxygen-70% nitrogen gas mixture at a rate of 120/min and a tidal volume of 2.0 ml while receiving 2.1 ml of saline per hour. The second, or low bicarbonate group, was hyperventilated with the 30% oxygen-70% nitrogen gas mixture by increasing the ventilatory rate to 140 min and the tidal volume to 2.25 ml. Simultaneously, the venous infusate was changed to 0.2 *N* HCl in saline given at approximately 2.1 ml/hr. The third, or high bicarbonate group, was ventilated with a 10% carbon dioxide, 30% oxygen, 60% nitrogen gas mixture at a rate of 120/min and a tidal volume of 2.0 ml. Simultaneously, the venous infusate was changed to a 0.35 M sodium bicarbonate solution given at approximately 2.1 ml/hr. The mean total amounts of sodium administered to the normal, low and high bicarbonate groups in the experimental period were 1.28, 1.28 and 1.46 mEq, respectively. In the low and high bicarbonate groups, 0.2-ml blood samples were also taken at 15-min intervals for the first hour of the experimental period to ascertain the constancy of the blood pH. This was not done in all animals once the procedure had been standardized.

Isohydric experiments in rats with bilateral ureteral ligation. At the conclusion of the control period, both ureters in each animal were ligated. The animals were then divided into three groups, normal, low and high bicarbonate and treated as in the previous isohydric experiments. In no instance was any urine excreted during the 150-min experimental period.

Chronic hypokalemia experiments. Male Sprague-Dawley rats weighted 350 to 450 g were placed on a zero potassium, zero chloride diet (obtained from Nutritional Biochemical Corporation, Cleveland, Ohio). Animals were allowed water containing 75 mEq/liter of sodium bicarbonate *ad lib* during the entire study. Rats were maintained on the potassium chloride-deficient diet for a 15-day period. On days 8 through 12 each rat received a daily dose of 2 mg of furosemide administered i.p. As in the previous experiments, after the control period rats were divided into three groups: unchanged, lowered and elevated bicarbonate. These were given the same fluid, and ventilated identically to the normal, low, and high bicarbonate groups previously described.

Acute hyperkalemia experiments. Male Sprague-

Dawley rats weighting 350 to 450 g were each tube fed twice on the day prior to the procedure and once again on the morning of the experiment. Each tube feeding consisted of 2 ml of a 1 mg/ml triamterene solution and 10 ml of a 5 mEq/liter potassium chloride solution in 5% dextrose and water. As in the hypokalemia experiments, rats were divided into unchanged, lowered and elevated bicarbonate groups. In these experiments, however, each i.v. administered solution contained 5 mEq of potassium chloride and the infusion rate of each solution during the experimental period was 6.0 ml/hr.

Analytic methods. Arterial pH and P_{CO_2} were measured using a blood microsystem (Radiometer BMS3 MK2) at 37°C. Bicarbonate was calculated from the pH and P_{CO_2} using an alignment nomogram (Siggard-Anderson). Plasma and urine potassium and sodium concentrations were determined on flame photometer (IL) with an internal lithium standard. Muscle potassium was determined as previously described [14].

Results

Effect of F_iO_2 on plasma potassium concentration and stability of the preparation. As animals were to be placed on a respirator employing different concentrations of carbon dioxide, it was necessary to ventilate each rat with the same F_iO_2 to eliminate any effect this variable might have on plasma potassium concentration. Two concentrations of oxygen, 100% and 30%, were chosen arbitrarily and their effect on plasma potassium concentration was compared to the effect of ventilating the animals with room air. Arterial pH, P_{CO_2} and bicarbonate concentrations were unaffected by changing from room air to either F_iO_2 ($P > 0.2$). However, in rats ventilated with 100% O_2, plasma potassium concentration rose significantly above control ($P < 0.01$) in the experimental period. Thus, plasma potassium concentration increased 0.45 mEq over the first 120 min of the experimental period and an additional 0.75 mEq over the next 30 min when rats were exposed to 100% O_2. By contrast, in the 30% O_2 and room air groups, no significant change in plasma potassium concentration occurred over the full 150 min of the experiment. Rats ventilated with a 30% oxygen gas mixture thus maintained both plasma potassium and arterial acid-base values constant for at least 150 min demonstrating stability of the preparation. All subsequent experiments, therefore, employed a 30% F_iO_2 in each animal throughout both control and experimental periods.

Reversibility of the preparation. It has previously been shown that induction of respiratory acidosis causes an increase in plasma potassium concentration [3, 4]. Respiratory acidosis can be rapidly induced or terminated so this maneuver was employed to determine the reversibility of the preparation. The results are shown in Fig. 1. In each of the five animals studied, ventilation with a 10% carbon dioxide gas mixture resulted in a drop in arterial pH. The decrease for the group was from 7.40 to 7.08 or 0.32 pH units. Simultaneously, as shown in the upper portion of the figure, plasma potassium concentration rose in each of the five animals, the mean rise over the 60-min experimental period being 0.95 mEq/liter. Changing the gas mixture to 30% oxygen, 70% nitrogen restored arterial pH to baseline in each animal. Similarly, plasma potassium concentration returned toward normal in each of the five animals though the mean for the group still remained 0.5 mEq higher than control. This value, however, is identical to the rise of 0.5 mEq/liter found in the 30% oxygen group over the same 150-min period. Thus, the preparation demonstrates both stability and reversibility.

Effect of changes in arterial pH at constant arterial bicarbonate concentration on plasma potassium concentration. To further substantiate the effect of blood pH on plasma potassium concentration when serum bicarbonate concentration is constant, respiratory acidosis was induced over a 150-min period. The results are shown on the first line of Table 1. In the six animals studied, the fall in arterial pH and the rise in P_{CO_2} in the experimental period were statistically significant ($P < 0.01$) while the slight decrease in the plasma bicarbonate concentration was not statistically significant ($P > 0.2$). The effect of respiratory

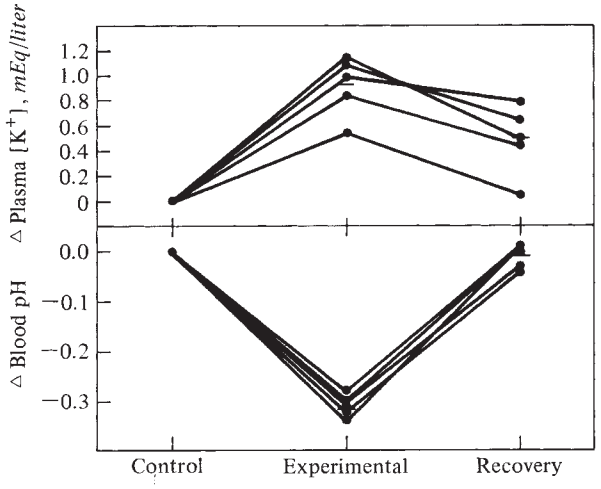

Fig. 1. *Reversibility of the preparation as demonstrated in acute respiratory acidosis.* Each *point* is the data obtained in a single experimental animal. *Horizontal bars* in the experimental and recovery time period represent the mean values for the group.

Table 1. Effect of arterial pH and bicarbonate concentration on plasma potassium[a]

	Arterial pH			P_{CO_2} *mm Hg*			Bicarbonate *mEq/liter*			Potassium *mEq/liter*		
	C^b	E_1	E_2	C^b	E_1	E_2	C^b	E_1	E_2	C^b	E_1	E_2
Respiratory acidosis ($N = 6$)	7.24 ±0.04	7.08[c,d] ±0.04	7.06[c,d] ±0.05	35.5 ±5.7	69.4[c,d] ±5.9	70.1[c,d] ±5.9	22.5 ±2.2	20.8 ±1.6	20.6 ±2.6	4.7 ±0.3	6.1[c,d] ±0.5	6.5[c,d] ±0.7
Normal bicarbonate ($N = 7$)	7.35 ±0.04	7.34 ±0.05	7.34 ±0.05	45.9 ±5.6	45.5 ±8.3	45.4 ±7.8	25.0 ±1.4	23.9 ±1.6	24.3 ±1.6	4.0 ±0.2	4.4 ±0.2	4.4 ±0.4
Low bicarbonate ($N = 10$)	7.36 ±0.05	7.35 ±0.07	7.33 ±0.05	38.0 ±6.3	23.3[c,d] ±4.3	19.8[c,d] ±5.9	21.0 ±2.4	12.8[c,d] ±3.0	10.9[c,d] ±2.8	4.3 ±0.6	5.2 ±0.7	6.7[c,d] ±0.8
High bicarbonate ($N = 7$)	7.39 ±0.02	7.37 ±0.05	7.40 ±0.03	35.9 ±4.1	81.4[c,d] ±8.3	74.7[c,d] ±5.9	21.5 ±1.5	44.1[c,d] ±5.6	44.3[c,d] ±5.6	4.3 ±0.3	4.6 ±0.6	4.6 ±0.6

[a] Each value represents the mean ± 1 SD.
[b] Represents the mean of the two control period values.
[c] Experimental period differs significantly from control period using Student's t test ($P < 0.01$).
[d] Differs significantly from experimental value of the normal group using Student's t test ($P < 0.01$).

acidosis on plasma potassium concentration is better seen in Fig. 2 where the change in plasma potassium concentration from control as a function of time is plotted. Data obtained in seven normal bicarbonate animals (detailed in Table 1) are plotted for comparison purposes. Plasma potassium concentration rose in the respiratory acidosis group, the rise, averaging 1.8 mEq at the conclusion of the 150-min experimental period in comparison to a rise of only 0.35 mEq/liter in the normal group. The difference between the two groups was statistically significant ($P < 0.001$).

Effect of changes in serum bicarbonate concentration on plasma potassium concentration under isohydric conditions. The first three columns of Table 1 show the extracellular acid-base conditions in the normal, low and high bicarbonate groups. It is apparent that isohydric conditions were maintained as arterial pH values during control and experimental periods did not differ by more than 0.03 pH units in any group. In both the low and high bicarbonate groups, the P_{CO_2} and the bicarbonate concentrations differed significantly between control and experimental periods ($P < 0.001$). In addition, plasma bicarbonate concentration and P_{CO_2} values differed significantly in the experimental period between each of the three groups ($P < 0.001$). As shown in the fourth column of Table 1, as well as in Fig. 3, the plasma potassium concentration rose significantly only in the low bicarbonate group ($P < 0.001$). Thus, after 150 min of experimental conditions, plasma potassium concentration had increased 2.4 mEq/liter in the low bicarbonate group, 0.4 mEq/liter in the normal bicarbonate group and 0.3 mEq in the high bicarbonate group. Urinary potassium excretion also differed in

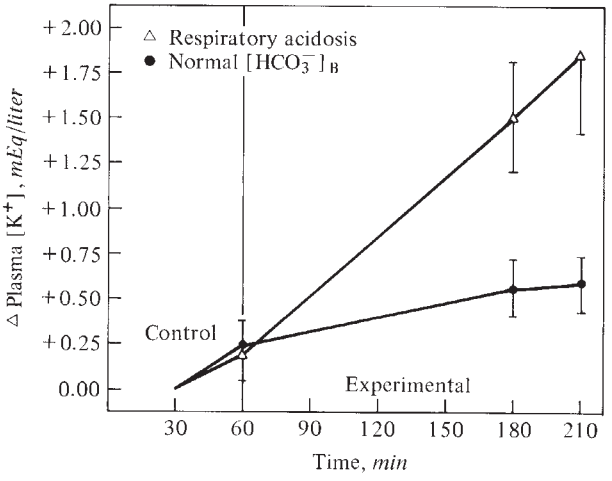

Fig. 2. *Effect of respiratory acidosis on plasma potassium concentration.* The *brackets* represent ± 1 SD. The *vertical line* at 60 min separates control and experimental periods.

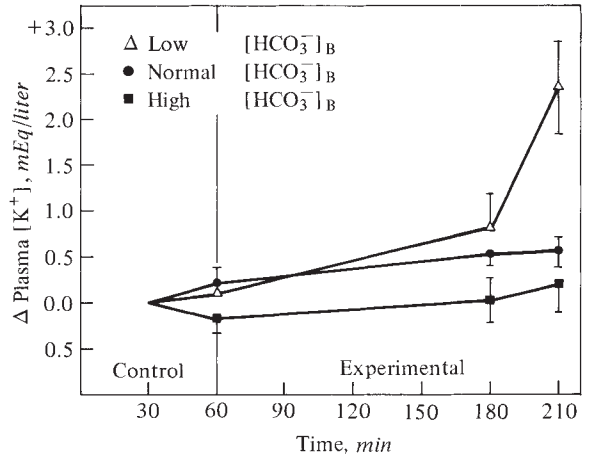

Fig. 3. *Effect on plasma potassium concentration caused by changing the blood bicarbonate concentration under isohydric external conditions.* The *brackets* represent ± 1 SD.

the three groups. During the experimental period, it rose above control in both the normal and high bicarbonate groups but was unchanged in the low bicarbonate rats. Mean potassium excretion for the experimental period was 0.037 mEq, 0.201 mEq and 0.373 mEq in the low, normal and high bicarbonate groups, respectively. Potassium excretion in the low bicarbonate group was significantly less than in either the normal or the high bicarbonate groups ($P < 0.001$).

Effect of changes in serum bicarbonate concentration on plasma potassium concentration under isohydric conditions in normokalemic rats with bilateral ureteral ligation. As potassium excretion in the normal and high bicarbonate rats during the experimental period is sufficient to account for the lesser increase in plasma potassium in these two groups compared to the low bicarbonate group, the previous experiments do not show whether bicarbonate affects the plasma potassium concentration by a nonrenal mechanism as well as a renal one. To study this problem experiments were performed in three groups of rats with ligated ureters to eliminate any urinary potassium loss. Table 2 shows the blood pH and bicarbonate in these groups. As in the previous experiments, arterial pH did not differ significantly between the control and experimental periods in any one of the three groups while in both low and high bicarbonate groups the P_{CO_2} and bicarbonate concentration differed significantly between the control and experimental periods ($P < 0.001$). Arterial P_{CO_2} and bicarbonate values also differed among all three groups in the experimental period ($P < 0.001$) while they did not differ within the control period ($P > 0.05$). Table 2 also shows the stability of bloood pH throughout the experimental period. Figure 4 shows the effect of these isohydric changes in bicarbonate concentration on plasma potassium in the rats with ligated ureters. After 120 min of the experimental period had elapsed, plasma potassium had risen 3.0 mEq in the low bicarbonate group, 1.75 mEq in the normal group and only 0.5 mEq in the high bicarbonate group. These values differed significantly from each other ($P < 0.01$). A large proportion of the low bicarbonate rats died shortly after this point in the experiment, presumably from hyperkalemia. Only rats in the normal and high bicarbonate groups, therefore, could be maintained for the full 150-min experimental period. It can be seen that over the final 30 min of the experimental period plasma potassium concentration continued to rise in the normal bicarbonate group while it remained constant in the high bicarbonate animals. Thus, the effect of bicarbo-

Table 2. Arterial acid-base conditions in the three isohydric groups of ureterally ligated rats[a]

	Arterial pH						Bicarbonate, *mEq/liter*					
	Control[b]	Experimental periods[c]					Control[b]	Experimental periods[c]				
		1	2	3	4	5		1	2	3	4	5
Normal bicarbonate ($N = 6$)	7.36 ±0.05	7.36 ±0.05	7.36 ±0.09	7.36 ±0.05	7.34 ±0.05	7.35 ±0.04	22.8 ±1.0	22.6 ±1.5	22.1 ±0.7	20.9 ±1.2	20.1 ±2.5	19.8 ±2.5
Low bicarbonate ($N = 6$)	7.32 ±0.06	7.37 ±0.06	7.35 ±0.03	7.32 ±0.03	7.30 ±0.07		24.6 ±1.8	20.4[d] ±1.7	18.4[d] ±1.2	16.5[d] ±1.9	11.3[d] ±4.0	
High bicarbonate ($N = 5$)	7.38 ±0.04	7.32 ±0.01	7.35 ±0.02	7.37 ±0.02	7.38 ±0.05	7.38 ±0.05	24.7 ±4.0	29.8 ±2.8	32.3[d] ±3.1	32.9[d] ±3.4	36.3[d] ±3.7	37.4[d] ±3.5

[a]Each value represents the ± 1 SD.
[b]Represents the mean of the two control period values as in Table 1.
[c]Experimental periods from 30 to 150 min at 30-min intervals.
[d]Experimental period differs significantly from control period ($P < 0.01$).

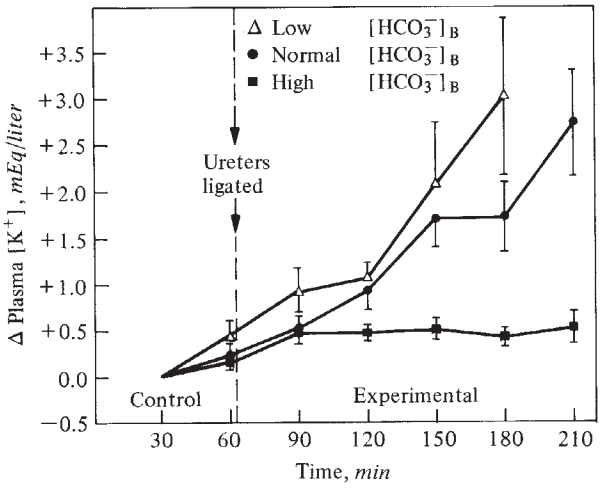

Fig. 4. *Effect on plasma potassium concentration of changing the blood bicarbonate concentration under isohydric external conditions in rats with ureteral ligation.* The *brackets* represent ± 1 SD.

nate on plasma potassium is demonstrable even when differences in urinary potassium loss are eliminated.

Effect of isohydric changes in blood bicarbonate concentration on plasma potassium in hypokalemia. Blood acid-base conditions in the three groups of hypokalemic animals are shown in Table 3. In the control period each of the groups exhibited a metabolic alkalosis. Differences in pH between any of the groups were nonsignificant despite a lower blood bicarbonate concentration in the unchanged bicarbonate group compared to the lowered bicarbonate group ($P > 0.05$). Experimental blood pH in any group did not differ significantly from control showing that the isohydric state was maintained. Bicarbonate concentration did, however, change significantly in both the lowered and elevated bicarbonate groups ($P < 0.001$). It was also altered significantly in the unchanged bicarbonate group ($P < 0.05$). The latter change can be ascribed to the effect of chloride administered to these potassium chloride-depleted animals.

Table 3 also shows that control period potassium values in each group were significantly reduced below normal. Differences in control plasma potassium concentration between groups were not significant. Experimental period plasma potassium concentration, however, was significantly altered from control in the lowered bicarbonate group ($P < 0.02$). The effect of bicarbonate in changing plasma potassium concentration in hypokalemia is better seen in Fig. 5 which depicts the change in plasma potassium from control as a function of time. At the end of the 150-min experimental period, differences between each of the groups were statistically significant. Thus, plasma potassium concentration had risen 1.2 mEq and 0.4

Table 3. Effect of bicarbonate concentration on plasma potassium concentration in potassium-depleted rats[a]

	pH			P_{CO_2} *mm Hg*			Bicarbonate *mEq/liter*			Potassium *mEq/liter*		
	C[b]	E_1	E_2	C[b]	E_1	E_2	C[b]	E_1	E_2	C[b]	E_1	E_2
Unchanged bicarbonate ($N = 9$)	7.53 ±0.09	7.52 ±0.09	7.52 ±0.09	38 ±11	35 ±9	33 ±8	31.1 ±3.3	28.1 ±3.6	27.0 ±2.9	1.8 ±0.4	2.1 ±0.5	2.2 ±0.4
Lowered bicarbonate ($N = 11$)	7.51 ±0.07	7.56 ±0.09	7.52 ±0.6	48 ±7	28[c] ±7	27[c] ±6	37.9 ±3.5	24.9[c] ±2.4	21.7[c] ±4.1	1.7 ±0.3	2.3 ±0.3	2.9[c] ±0.8
Elevated bicarbonate	7.51 ±0.04	7.45 ±0.06	7.46 ±0.05	42 ±4	84[c] ±9	84[c] ±6	34.2 ±1.3	53.8[c] ±6.9	59.8[c] ±3.1	2.0 ±0.4	2.0 ±0.3	1.8 ±0.2

[a]Each value represents the mean ± 1 SD.
[b]The mean of the two values obtained during the control period.
[c]Differs significantly from the control period value using the Student's *t* test ($P < 0.05$).

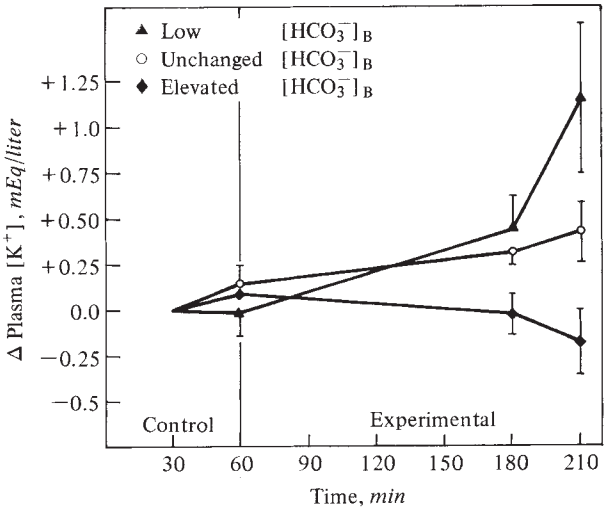

Fig. 5. *Effect of isohydric changes in blood bicarbonate concentration on the plasma potassium concentration in chronic hypokalemic rats.* The *brackets* represent ± 1 SD. The *vertical line* at 60 min separates control and experimental periods.

mEq in the lowered bicarbonate and unchanged bicarbonate groups, respectively, while it had fallen by 0.2 mEq in the elevated bicarbonate groups.

Figure 6 shows that these differences were not due to alterations in the urinary excretion of potassium. Urinary potassium excretion was low in all groups and did not rise significantly during the experimental period. Although urinary potassium excretion was highest in the low bicarbonate group, these were the animals who exhibited the greatest increase in their plasma potassium concentration. Even in this group absolute potassium excretion during the experimental period was extremely low being only 0.04 mEq while it was 0.01 mEq and 0.02 mEq in the unchanged and elevated bicarbonate groups, respectively.

Effect of isohydric changes in blood bicarbonate concentration on plasma potassium in hyperkalemia. Blood acid-base conditions in the three groups of hyperkalemic animals are shown in Table 4. Arterial pH did not differ significantly between groups in either the control or experimental periods ($P > 0.2$). Also, in no group did arterial pH in the experimental period differ significantly from control period values ($P > 0.4$) showing that isohydric conditions were maintained throughout the experiment in each group. Despite the statistical equality of arterial P_{CO_2} and bicarbonate concentrations in the three groups during the control period, these indices differed significantly between each group in the experimental period ($P < 0.001$) demonstrating a complete separation of the three isohydric groups. In the control period plasma potassium concentration was significantly above normal in all three groups of animals compared to values obtained in normokalemic rats ($P < 0.001$). Control plasma potassium concentration in the lowered bicarbonate group did not differ significantly from the control plasma potassium in the elevated bicarbonate group ($P > 0.05$) but did significantly exceed the value found in the unchanged bicarbonate group ($P < 0.05$). In both the unchanged and elevated bicarbonate groups, plasma potassium concentration fell during the experimental period ($P < 0.005$ and < 0.001, respectively) but did not change significantly in the lowered bicarbonate group ($P > 0.5$).

The effect of bicarbonate concentration in altering plasma potassium concentration in the hyperkalemic state is shown in Fig. 7. After either 120 or 150 min of experimental conditions, the change in plasma potassium concentration was significantly different among each of the three groups of animals. At the conclusion of the experimental period, plasma potassium concentration had risen 0.1 mEq in the lowered bicarbonate group, while in the unchanged and elevated bicarbonate groups it had decreased by 1.3 and 2.0 mEq, respectively.

As demonstrated in hypokalemia, differences in plasma potassium concentration among groups were not caused by differences in urinary potassium excretion during the experimental period. This is shown in Fig. 8. Although urinary potassium excretion rose in the experimental period in all three groups, there were no statistically significant differences among them. Absolute potassium excretion in the experimental period in the lowered, unchanged and elevated bicarbonate groups was 0.7, 0.9, and 0.8 mEq, respectively. In addition, the four animals in the elevated bicarbonate group who decreased

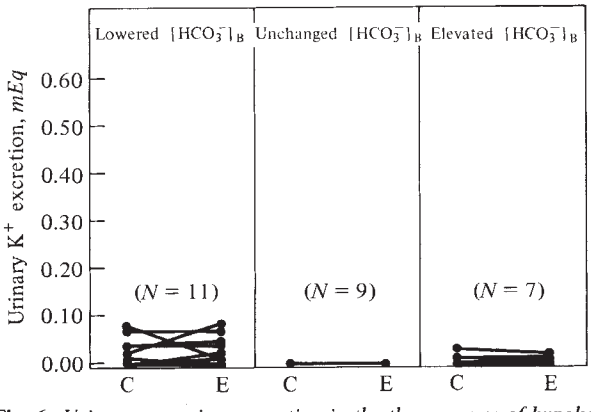

Fig. 6. *Urinary potassium excretion in the three groups of hypokalemic rats during control (C) and experimental (E) periods.* Each *line* represents data obtained from a single rat. The number of rats in each group is given in parenthesis within the group since many of the rats have identical values.

Table 4. Effect of bicarbonate concentration on plasma potassium concentration in acutely hyperkalemic rats[a]

	pH			Pco_2 *mm Hg*			Bicarbonate *mEq/liter*			Potassium *mEq/liter*		
	C[b]	E_1	E_2	C[b]	E_1	E_2	C[b]	E_1	E_2	C[b]	E_1	E_2
Unchanged bicarbonate ($N = 6$)	7.27 ±0.04	7.27 ±0.06	7.27 ±0.05	47 ±3	45 ±3	44 ±7	21.4 ±2.0	20.3 ±1.8	19.9 ±2.4	6.6 ±0.4	5.4[c] ±0.3	5.3[c] ±0.4
Lowered bicarbonate ($N = 7$)	7.28 ±0.05	7.25 ±0.07	7.27 ±0.05	51 ±7	34[c] ±4	30[c] ±3	23.5 ±0.8	14.4[c] ±1.4	13.4[c] ±1.1	7.5 ±0.4	7.4 ±0.3	7.6 ±0.2
Elevated bicarbonate ($N = 6$)	7.29 ±0.05	7.29 ±0.05	7.31 ±0.03	45 ±10	85[c] ±15	88[c] ±14	20.7 ±2.7	40.3[c] ±4.1	43.9[c] ±5.4	6.9 ±0.3	4.9[c] ±0.2	4.9[c] ±0.3

[a] Each value represents the mean ± 1 SD.
[b] The mean of the two values obtained during the control period.
[c] Differs significantly from the control period value using Student's t test ($P < 0.05$).

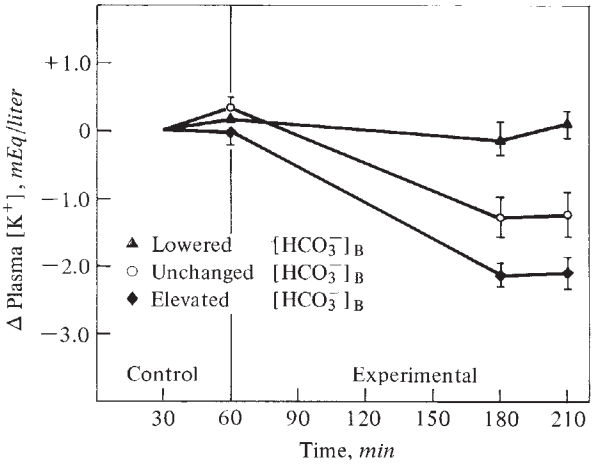

Fig. 7. *Effect of isohydric changes in blood bicarbonate concentration on the plasma potassium concentration in acutely hyperkalemic rats.* The *brackets* represent ± 1 SD.

potassium excretion during the experimental period lowered their plasma potassium concentration to an extent equal to that of the two animals in whom urinary potassium excretion increased.

Table 5 shows the tissue potassium level in the hypokalemic and hyperkalemic experiments. The only significant effect of changing bicarbonate was found in hypokalemia where lowered bicarbonate muscle potassium was decreased ($P < 0.05$) below that in the elevated bicarbonate animals. Muscle potassium content in all three groups of rats with hyperkalemia was higher than muscle potassium content found in either the normokalemic or hypokalemic animals.

Discussion

Hyperkalemia is often found in clinical disorders involving extracellular acidosis, while hypokalemia is seen in alkalotic conditions [5, 15, 16]. Fenn and Cobb [2], as early as 1934, postulated that pH is the major factor regulating serum potassium concentration. This hypothesis was substantiated by Scribner, Fremont-Smith and Burnell [4] who demonstrated in dogs that acute respiratory acidosis, unaccompanied by significant change in bicarbonate concentration, raised serum potassium concentration and prevented infused potassium from entering cells. This phenomenon was not restricted to animals as studies in humans showed that either metabolic or respiratory acidosis raised serum potassium concentration [5]. In the absence of significant urinary potassium loss, it was reasonable to assume that lowering of the extracellular pH caused a shift of potassium from the intracellular to the extracellular compartment. Direct

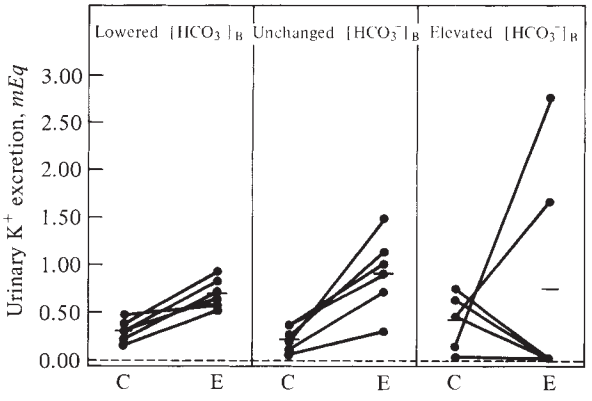

Fig. 8. *Urinary potassium excretion in the three groups of hyperkalemic rats during control (C) and experimental (E) periods.* The control point represents the total urinary potassium excretion during the 60-min control period while the experimental point represents total urinary potassium excretion during the entire 150-min experimental period.

experimental confirmation was given by Mudge and Vislocky [17], who reported decreased intracellular potassium in human striated muscle in metabolic acidosis.

Contrary to the unequivocal role extracellular pH plays in regulating serum potassium concentration and the intracellular to extracellular potassium ratio is the regulatory role exerted by bicarbonate in these same processes. Liebman, Birkenfield and Edelman [11], for example, found a greater correlation between serum potassium concentration and extra-cellular pH in metabolic than in respiratory acid-base disorders. These workers concluded that bicarbonate was important in the regulation of serum potassium concentration but definitive data were not presented. Makoff, DaSilva and Rosenbaum [13] expanded the extracellular volume of nephrectomized dogs hypertonically and demonstrated changes in extracellular potassium which did not correlate with changes in extracellular pH but did appear to correlate with changes in extracellular bicarbonate. They felt that bicarbonate was a major regulator of extracellular potassium concentration. In addition, Kim and Brown [12] using a nephrectomized dog model found that under isohydric external conditions lowering the blood bicarbonate concentration raised the serum potassium concentration. In their experiments, however, administration of normal saline also significantly raised this value. Simmons and Avedon [10], moreover, reported that changes in the extracellular bicarbonate concentration had no effect on serum potassium in dogs maintained under isohydric external conditions. There was, however, much scattering of their data and the time of the experiment was relatively brief.

Data from the present study support the concept that plasma bicarbonate concentration directly regulates the plasma potassium concentration. Two mechanisms seem to be operative. The first, a renal one, is probably due to changes in delivery of bicarbonate to the distal nephron. Increased delivery of the relatively nonreabsorbable bicarbonate ion to the distal site should enhance potassium ion secretion into the lumen [18]. The second mechanism, demonstrated directly in the animals with bilateral ureteral ligation and indirectly in those with hypokalemia or hyperkalemia, indicates that bicarbonate also affects the transcellular distribution of potassium. In the ureteral ligation experiments, urinary potassium loss was prevented, while in the hypokalemia and hyperkalemia experiments the urinary potassium excretion data cannot account for the results. Only in the chronic hypokalemia experiments, however, was a significant change in muscle potassium content recorded. The inability to demonstrate changes in muscle potassium content in normokalemia and hyperkalemia does not mitigate against the hypothesis that bicarbonate moves potassium into the cell. In the normal rat, extracellular potassium represents only 1 to 2% of the total body potassium and even large shifts of extracellular potassium will alter tissue potassium very little. Indeed, the expected change is well within the error of measurement given the variability of muscle potassium between animals

Table 5. Effect of bicarbonate concentration on tissue potassium content in hypokalemic and hyperkalemic rats[a]

	Hypokalemia			Hyperkalemia		
	Unchanged	Lowered	Elevated	Unchanged	Lowered	Elevated
Muscle potassium *mEq/kg of dry wt*	328.2 ±21.5	291.1 ±28.0	343.4[b] ±22.1	488.5 ±22.5	488.2 ±32.9	493.9 ±24.1
Kidney potassium *mEq)kg of dry wt*	286.9 ±29.4	273.6 ±33.7	300.4 ±19.1	332.8 ±34.7	337.4 ±29.1	320.9 ±16.8

[a]Each value represents the mean ± 1 SD.
[b]Differs significantly from the value of the lowered bicarbonate group using Student's *t* test ($P < 0.05$).

and the measurement technique employed. The demonstration of a significant change in muscle potassium in the hypokalemia experiments might be fortuitous or could be due to the 40% reduction in muscle potassium. Even in these experiments, however, significance was achieved only at the 5% probability level.

Although these experiments did not investigate the means by which bicarbonate alters transcellular potassium movement, the results support the concept that mammalian muscle cells are permeable to external bicarbonate ions. Adler, Roy and Relman [19, 20] previously showed *in vitro* that extracellular bicarbonate affects intracellular pH but the experiments were unable to distinguish between the movement of hydrogen ion out of the cell and hydroxyl or bicarbonate movement in the opposite direction. In the present experiments, extracellular pH was held constant making it appear that bicarbonate movement was responsible for the observed changes in plasma potassium. As the muscle membrane is permeable to bicarbonate, the distribution of this ion between the extracellular and intracellular compartments should be similar to that of chloride, another monovalent anion. Yet, the extracellular to intracellular chloride ratio in muscle appears to be at least ten times greater than that of bicarbonate [21] indicating active transport of bicarbonate into mammalian muscle cells. It should be noted that in all the present experiments the changes in blood bicarbonate concentration were accompanied by inverse changes in serum chloride. A role for extracellular chloride in regulating transcellular potassium distribution cannot, therefore, be ruled out.

The present experiments also call attention to the growing body of data showing that bicarbonate ions, separate from their effect on extracellular pH, alter and regulate metabolic activity. Isohydric changes in extracellular bicarbonate concentration have previously been shown to alter lactate production [6], glycogen synthesis in the liver [22], renal gluconeogenesis [8] and muscle cell pH heterogeneity [23]. Plasma potassium concentration must now be added to this growing list. The results emphasize the importance of controlling both pH and bicarbonate concentration in biological experiments.

Finally, extrapolation of these *in vivo* rat studies to the human could have important clinical implications. Most reviews and case studies in patients with hyperkalemia have shown that raising blood pH decreases serum potassium concentration. None of these studies, however, discuss the role of bicarbonate separate from its effect upon the blood pH [24–27]. At least two reviews [28, 29] do mention that bicarbonate is effective in treating hyperkalemia even when blood pH is compensated but no data are given to support this conclusion nor is the mechanism of action discussed. Furthermore, a search of the literature fails to reveal instances in which hyperkalemia has been treated by bicarbonate in patients with compensated acid-base disorders. The present results indicate that administration of bicarbonate to patients with hyperkalemia will lower the plasma potassium concentration rapidly even when the blood pH is within normal limits. Since this effect is independent of renal function, bicarbonate administration to patients with hyperkalemia should be effective even if renal function is impaired or absent. Obviously, studies in humans will have to be performed to corroborate this conclusion.

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Reprint requests to Dr. Sheldon Adler, Montefiore Hospital, 3459 Fifth Avenue, Pittsburgh, Pennsylvania 15213, U. S. A.

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